NTP
National Toxicology Program

The National Toxicology Program (NTP) High Throughput Screening (HTS) Initiative

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NTP/NIEHS

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- **NTP Mission (http://ntp.niehs.nih.gov/):**
  - Coordinate toxicology testing programs within the federal government.
  - Strengthen the science base in toxicology.
  - Develop and validate improved testing methods.
  - Provide information about potentially toxic chemicals to health, regulatory, and research agencies, scientific and medical communities, and the public.

- **NTP Bioassay:**
  - Compounds are nominated for testing using a transparent process; fewer than 20 are tested each year.
  - Testing plan: (1) mutagenicity (2) sub-chronic toxicity (3) two-year carcinogenicity.
  - Specialty testing can be conducted in immunotoxicity, developmental / reproductive toxicity, genetic toxicity.

To meet the challenge of 21st century toxicology, the NTP Roadmap includes a major initiative to develop a high throughput screening (HTS) program with 3 main goals:

- To prioritize chemicals for further in-depth toxicological evaluation
- To identify mechanisms of action
- To develop predictive models for *in vivo* biological response

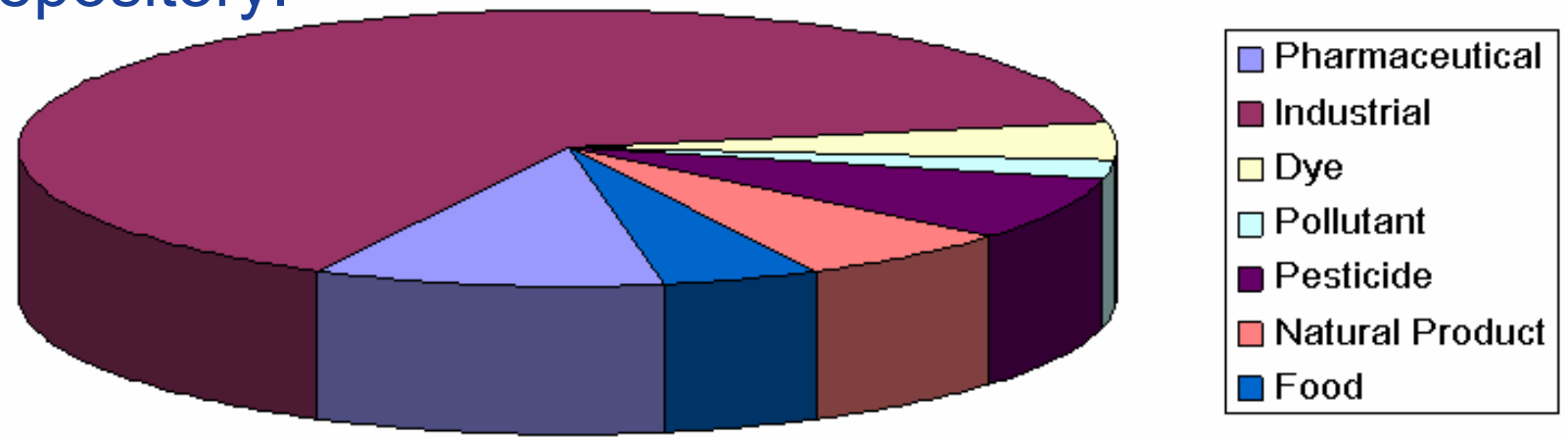
NIH Molecular Libraries Initiative

http://nihroadmap.nih.gov/molecularlibraries/

- HTS methods are being used to identify small molecules that can be optimized as chemical probes to study the functions of genes, cells, & biochemical pathways.
- In mid-2005, NTP became a formal participant in the MLI by establishing a collaboration with the NIH Chemical Genomics Center (NCGC)
- As a result, the NTP has the opportunity to link data generated from HTS assays for biological activity to toxicity data produced by the NTP's testing program.

The first 1408 Compound Set

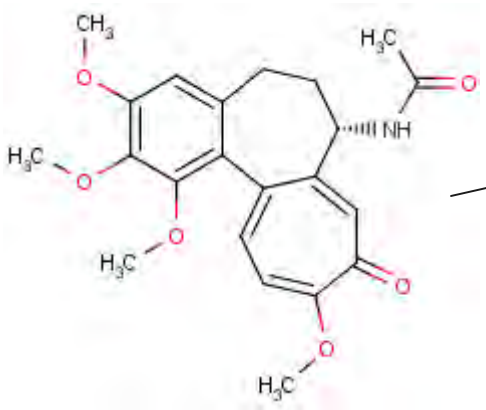
- 55 duplicates, 1206 with NTP data, 147 are reference substances for the validation of alternative *in vitro* test methods (e.g., dermal corrosion, acute toxicity, endocrine activity).
- A subset of these compounds will be made available to all MLI Centers through the MLI Chemical Repository.



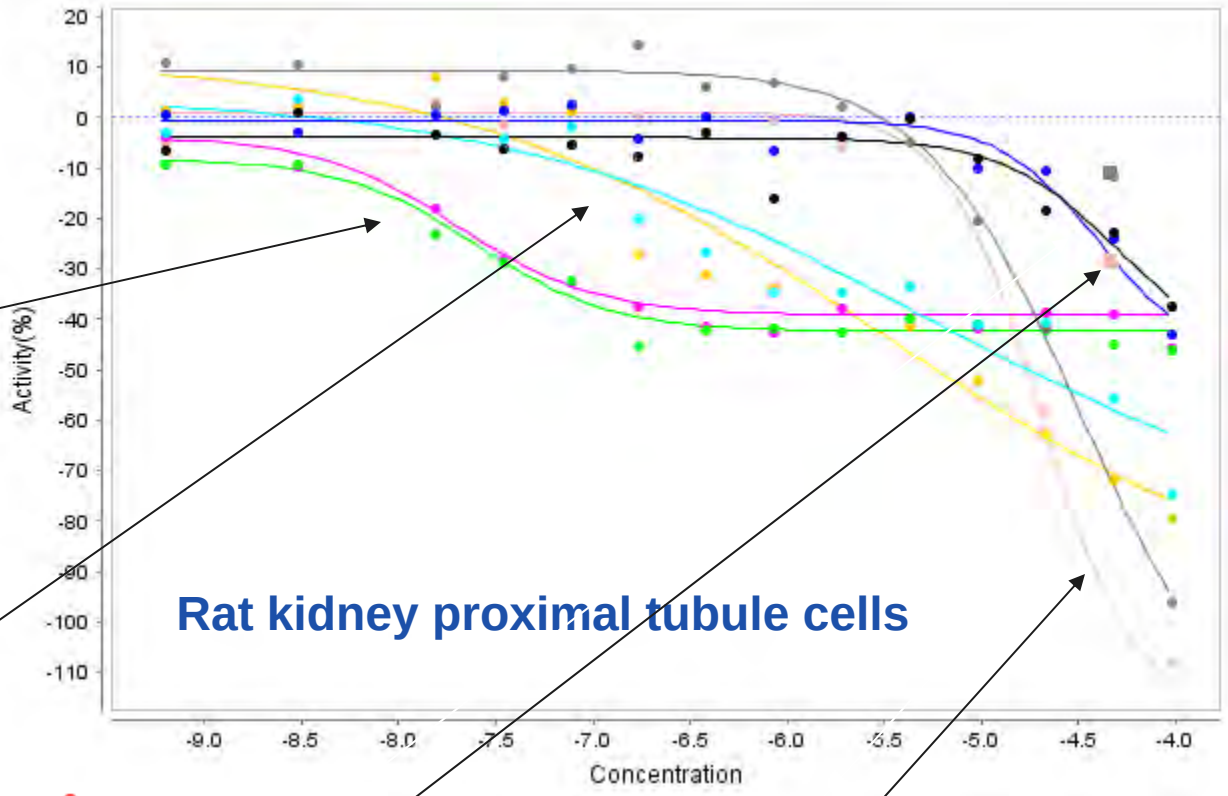
Assays supplied to the NCGC

- Cytotoxicity Assays
  - CellTiter-Glo® Luminescent Cell Viability Assay (measures ATP levels)
  - Cytotox-ONE™Homogeneous Membrane Integrity Assay (measures LDH release)
- Apoptosis Assays
  - Caspase-Glo® 3/7 Assay
  - Caspase-Glo® 9 Assay
  - Caspase-Glo® 8 Assay
- P-glycoprotein (Pgp) ATPase Assay (aka MDR1 or ABCB1)
  - Pgp-Glo™ Assay

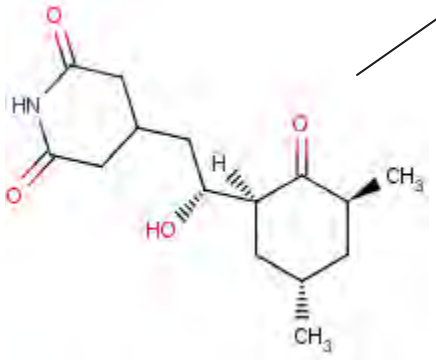
Assay Reproducibility:



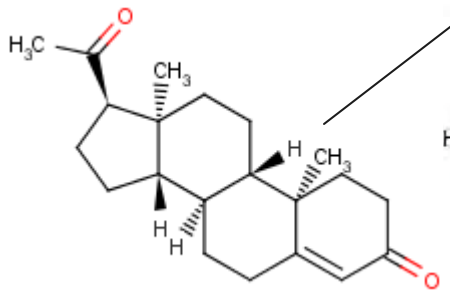
Colchicine



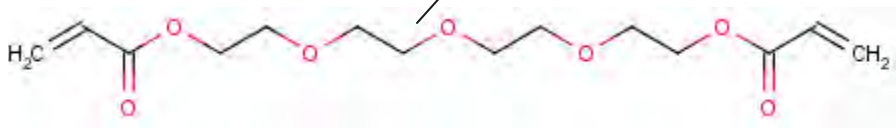
Rat kidney proximal tubule cells



Cycloheximide

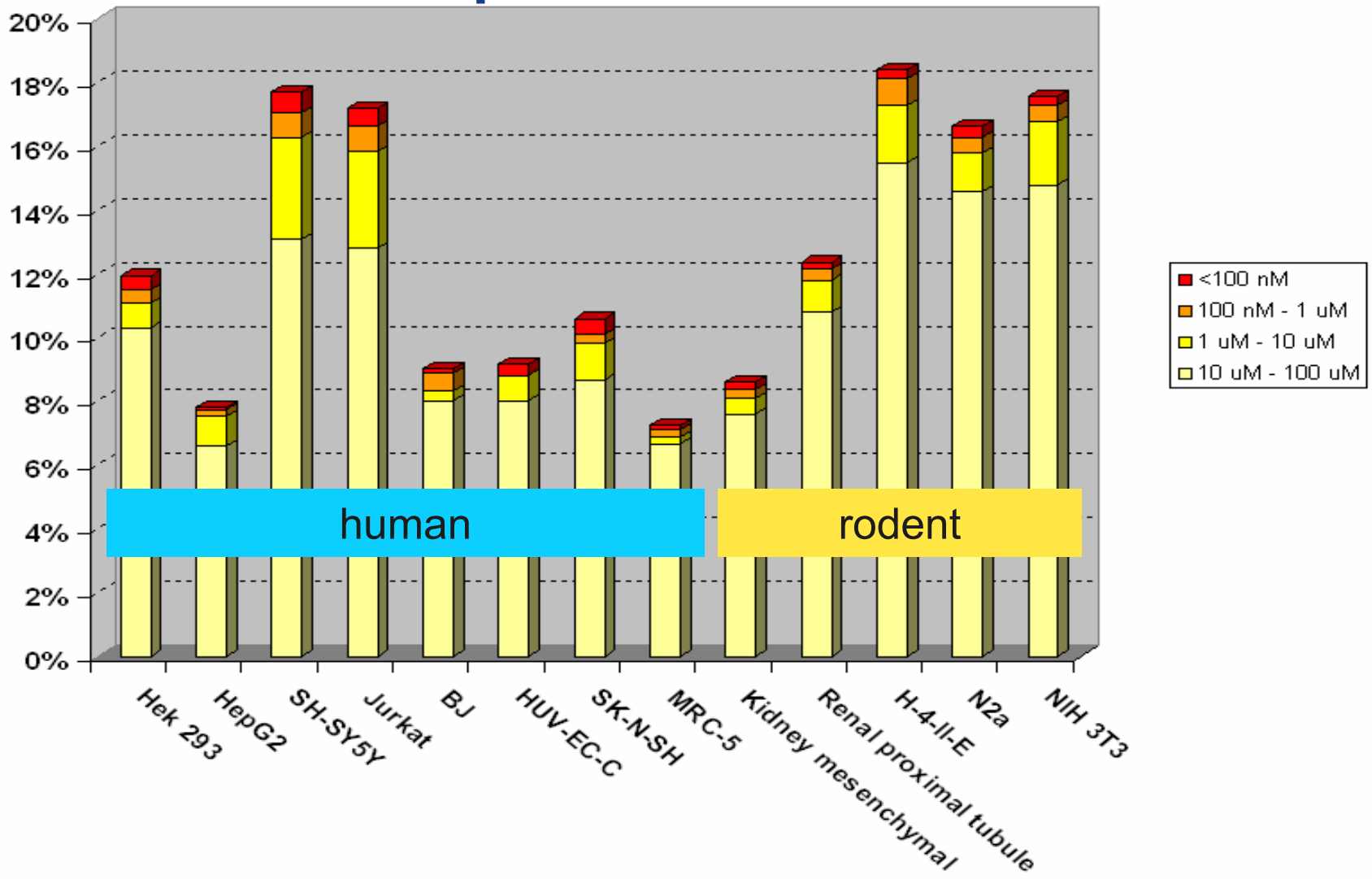


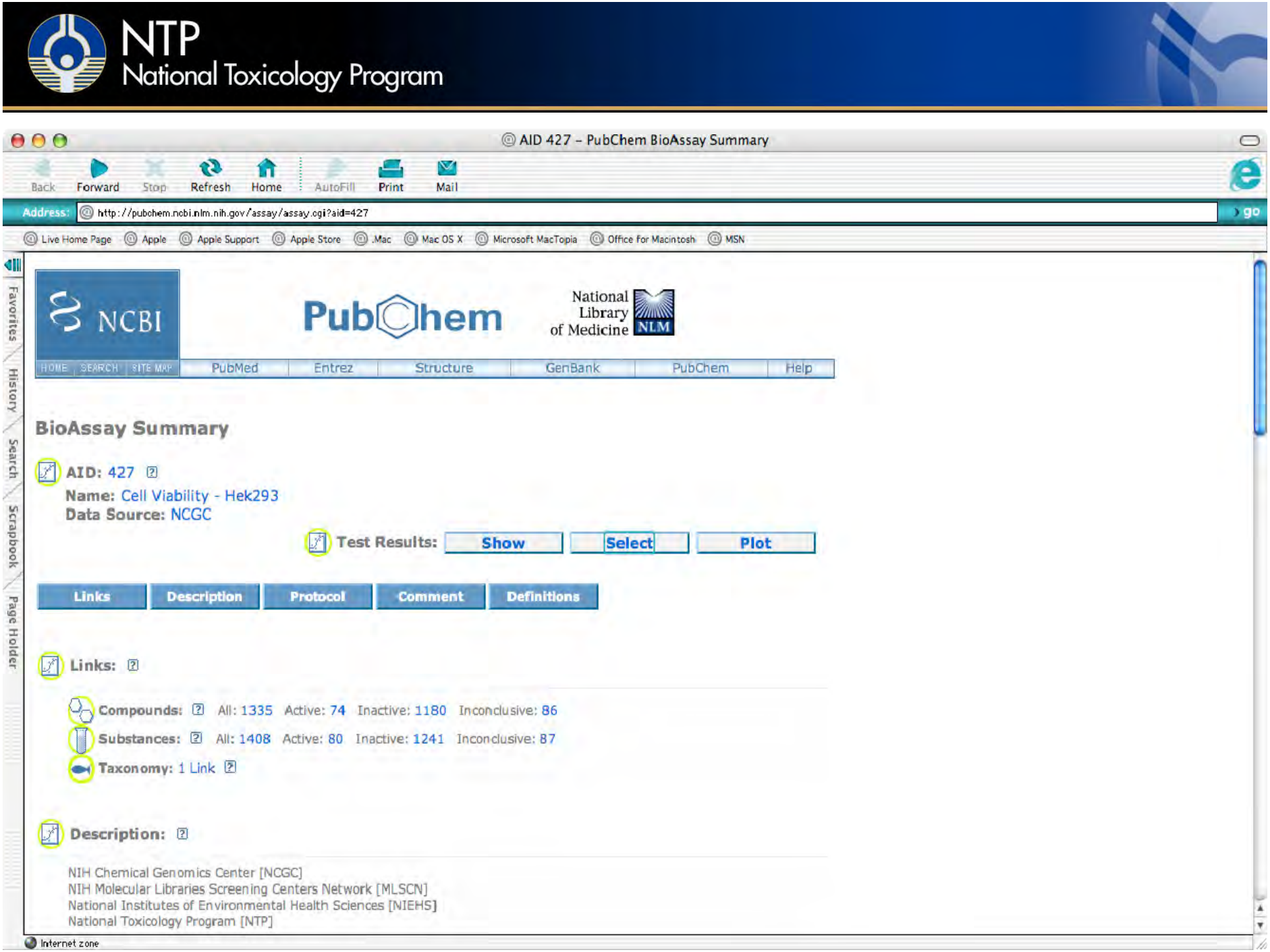
Progesterone



Tetraethylene Glycol Diacrylate

Cytotoxicity potency distribution of the NTP 1408 compounds in 13 cell lines

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Microsoft MacTopia
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MSN
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Structure
GenBank
PubChem
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Data Source: NCGC
Test Results:
Show
Select
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Description
Protocol
Comment
Definitions
Links:
Compounds: All: 1335 Active: 74 Inactive: 1180 Inconclusive: 86
Substances: All: 1408 Active: 80 Inactive: 1241 Inconclusive: 87
Taxonomy: 1 Link
Description:
NIH Chemical Genomics Center [NCGC]
NIH Molecular Libraries Screening Centers Network [MLSCN]
National Institutes of Environmental Health Sciences [NIEHS]
National Toxicology Program [NTP]
Internet zone

NTP National Toxicology Program

Future Directions

- Provide a second set of 1408 to the NCGC that will include structurally-related compounds with a range of activities in immunotoxicity and carcinogenicity.
- Expand the number of HTS assays for critical pathways of interest by including selected assays available at the MLI centers and the via the EPA ToxCast assays.
- Expand the number of cell types by including primary cells.
- Expand bioinformatics/data mining capabilities within NTP and across publicly accessible databases.

NTP/NIEHS

- John Bucher, Allan Dearry, Jennifer Fostel. Chris Portier, Fred Parham, Cynthia Smith, Kristine Witt

NCGC

- Chris Austin, Ruilli Huang, Jim Inglese, Noel Southall, Menghang Xia

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